

## 29th Plant Development Workshop

Dear Colleague,

I am enclosing the schedule for the 29th Plant Development Workshop to be held here at the University of Toronto on Saturday, March 26, 1994. We have an interesting program for the day, one that includes a diversity of approaches to plant development.

Registration, morning coffee, and oral presentations will be held in the Koffler Institute of Pharmacy Management, 575 Spadina Avenue. Lunch and the poster session will be held in the lobby of the Earth Sciences Centre, 5 Bancroft Avenue. A map of the U of T campus with these locations marked is enclosed. There is no registration fee, but there will be a \$5 charge for a hot lunch. If possible, bring a mug for coffee and juice.

For those of you driving to the Workshop, our location is near the intersection of Spadina Avenue and College Street. Parking is available behind the Koffler Institute (Lot #19 on the enclosed map) and at 1 Spadina Crescent (Lot #10). For both lots, you need to have **\$3.75 in change**, obtain a ticket from the meter in Lot #19, and leave the ticket on the dashboard. If you are taking the TTC, come to the Spadina Station (bus #77 or 10 min walk south) or to the Queens Park Station (westbound streetcar or 10 min walk west).

The length of oral presentations (plus discussion) is 15 minutes. If you are presenting a poster, the poster boards will be set up in the Earth Sciences Centre lobby from 8:30 am. You may want to set up your poster before the talks start at 9:30 am. There will be enough space for each poster to be up to eight feet wide and four feet high. These poster boards use a stick-on velcro system, and we will have supplies available.

We are looking forward to seeing you on March 26th!



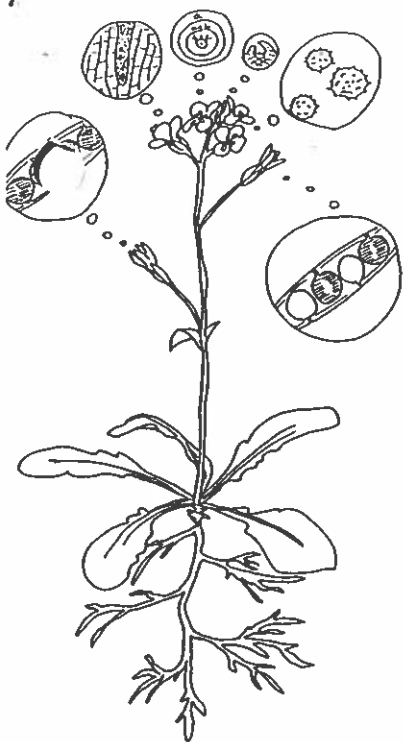
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## 29th PLANT DEVELOPMENT WORKSHOP

**Saturday, March 26, 1994**

**Department of Botany**

**University of Toronto**

### **PROGRAM**

- 9:00 Registration and Coffee - Koffler Institute
- 9:25 Welcoming remarks - NANCY G. DENGLER
- 9:30 RODGER C. EVANS and TIMOTHY A. DICKINSON, Department of Botany, University of Toronto and Royal Ontario Museum. Floral development of meristic variants in Crataegus sect. Douglasii Lindley (Rosaceae: Maloideae).
- 9:45 CHRISTINE M. KAMPNY, Department of Botany, University of Toronto and Royal Ontario Museum. Variation in corolla development in the Veroniceae.
- 10:00 JUNKO KITAGAWA and USHER POSLUSZNY, Department of Botany, University of Guelph. Preliminary study of inflorescence variations of Lilaea scilloides.
- 10:15 TAMMY L. SAGE, Department of Botany, University of Toronto. Ovarian self-incompatibility in Asclepias exaltata.
- 10:30 C. HASENKAMPF, N. JAYAWARDENE, M. QURESHI and M. DOOKHERAN, University of Toronto, Scarborough Campus. Chromosomal distribution of meiotic molecules.
- 10:45 Coffee Break

- 11:00 JEAN M. GERRATH and NANCY G. DENGLER, Department of Horticultural Science, University of Guelph and Department of Botany, University of Toronto. Primary vascular patterns in the Vitaceae.
- 11:15 JANE P. YOUNG, Department of Botany, University of Toronto. The developmental basis of heterophylly in Ranunculus flabellaris.
- 11:30 DARYL E. ENSTONE and CAROL A. PETERSON, Department of Biology, University of Waterloo. Band plasmolysis and the development of suberin lamellae in the exodermis of Zea mays L. roots.
- 11:45 A. DARLINGTON and J. DAT, Faculty of Forestry, University of Toronto. Root growth under water stress.
- 12:00 JANICE CHRISTIAN and ALICJA ZOBEL, Department of Chemistry, Trent University, Peterborough. Competition of pine seedlings and grass under water stress.
- 12:15 Lunch and Poster Session - Earth Sciences Centre Lobby
- 2:15 C. DANIEL RIGGS and ANDREA HORSCH, Department of Botany and Centre for Plant Biotechnology, University of Toronto, Scarborough College. Utilities of a polyclonal antiserum directed against DNA binding proteins from meiotic cells.
- 2:30 KALLIE KEITH and PETER McCOURT, Department of Botany, University of Toronto. Fusca 3: A heterochronic mutation affecting late embryo development in Arabidopsis.
- 2:45 SEAN CUTTLER, Department of Botany, University of Toronto. ABA supersensitive mutants in Arabidopsis.
- 3:15 KEYNOTE TALK: DR. PETER McCOURT, Department of Botany, University of Toronto. Developing a genetic approach to the study of plant development.
- 4:15 Reception (refreshments)

## POSTERS

CHERYL ASHBY and ALICJA ZOBEL, Department of Chemistry, Trent University, Peterborough. Possible use of scopoletin and imperatorin as anticancer agents.

E. FOSTER ATKINSON and JUDY N. STROMMER, Department of Molecular Biology and Genetics, University of Guelph. Alcohol Dehydrogenase 1 encodes the anther-predominant ADH polypeptide from Petunia "Mitchell".

D. BARABÉ, Institut de Recherche en Biologie Végétale, Montréal. The concepts of open system and closed system in phyllotaxis.

PENNY E. BEECROFT and JOHN N.A. LOTT, Department of Biology, McMaster University, Hamilton. Leakage of mineral nutrients from imbibing pollen grains.

G. CLOUTIER, J. DAT and A. DARLINGTON. Faculty of Forestry, University of Toronto. Humidity and the hydraulic conductance of Phaseolus vulgaris.

MICHELLE DOOKHERAN and C.A. HASENKAMPF. University of Toronto, Scarborough Campus. In situ hybridization with lily meiotic chromosomes.

JEAN M. GERRATH, JOAN E. KROCHKO and JUDY N. STROMMER, Department of Horticultural Science, University of Guelph. Development of leaf tendrils in pea: a progress report.

JOHN N.A. LOTT, M. MARCIA WEST, BEN CLARK and PENNY E. BEECROFT. Department of Biology, McMaster University, Hamilton. Changes in globoid crystal composition in castor bean cotyledons and endosperm during early seedling growth.

MISBAH QURESHI, University of Toronto, Scarborough Campus. An immunocytochemical investigation to understand the biological function of meiotin-1 in Lilium longiflorum.

C. DANIEL RIGGS. Department of Botany and Centre for Plant Biotechnology, University of Toronto, Scarborough Campus. Molecular cloning of cDNAs encoding meiotin-1: a meiotic protein associated with strings of nucleosomes.

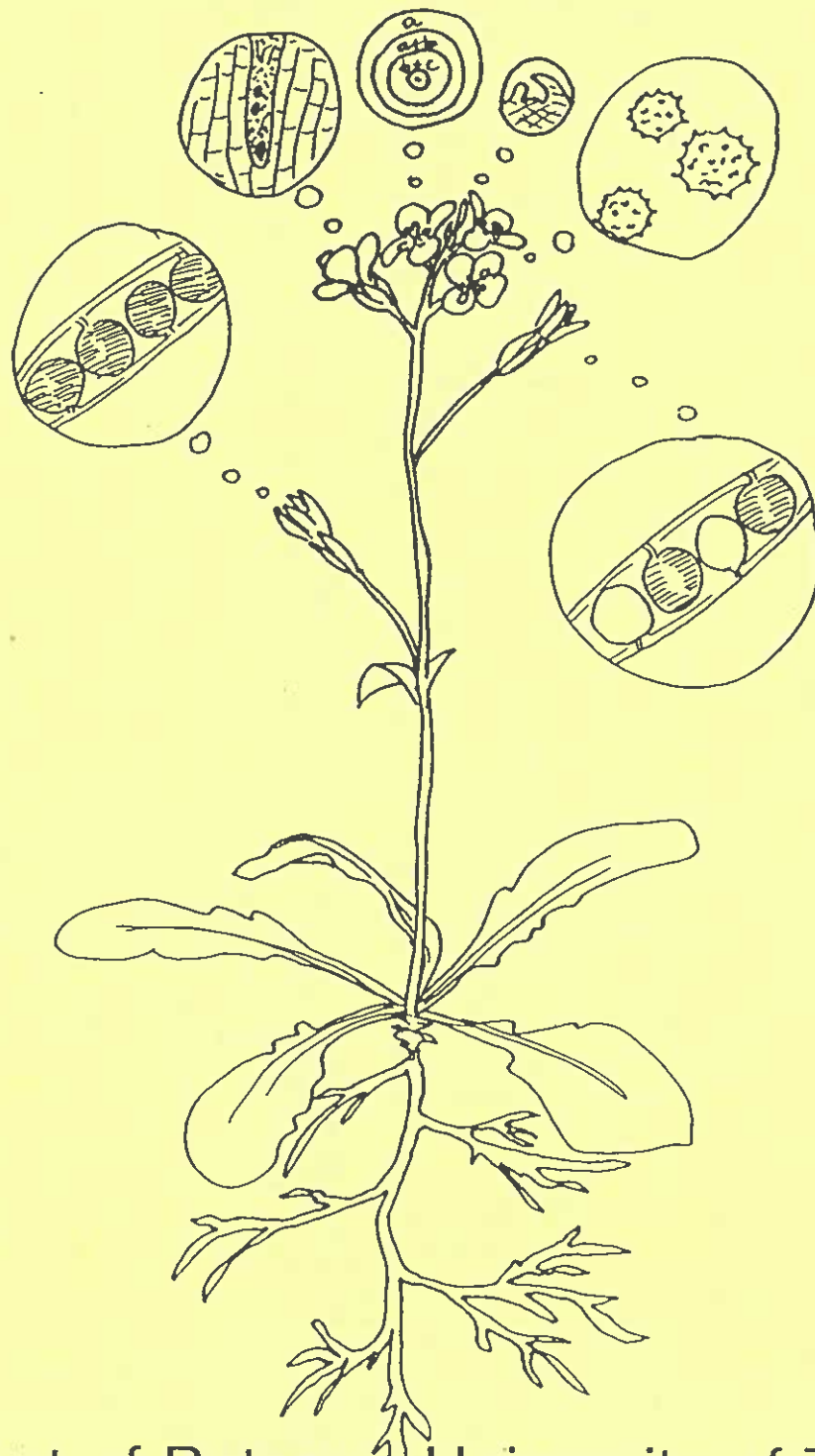
S.E. RIVERS, P.S. SUMMERS and E.A. WERETILNYK, Department of Biology, McMaster University, Hamilton. Serine decarboxylase from spinach.

D.D. SMITH, P.S. SUMMERS and E.A. WERETILNYK, Department of biology, McMaster University, Hamilton. Light and salt regulation of a choline biosynthetic enzyme in spinach: s-adenosyl-l-methionine: phosphoethanolamine n-methyltransferase.

G. THIBEAULT, D. BARABÉ and L. BROUILLET, Institut de Recherche en Biologie Végétale, Montréal. Development of asymmetry of the leaf of Bignoniaceae seedlings.



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Department of Botany University of Toronto

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MICHELLE DOOKHERAN and C.A. HASENKAMPF. University of Toronto, Scarborough Campus. In situ hybridization with lily meiotic chromosomes.

I. ELHASSANI, G-J GAO and A. NASSUTH. Department of Botany, University of Guelph. The expression and movement of Wheat Streak Mosaic Virus Coat Protein in infected wheat.

JEAN M. GERRATH, JOAN E. KROCHKO and JUDY N. STROMMER, Department of Horticultural Science, University of Guelph. Development of leaf tendrils in pea: a progress report.

JOHN N.A. LOTT, M. MARCIA WEST, BEN CLARK and PENNY E. BEECROFT. Department of Biology, McMaster University, Hamilton. Changes in globoid crystal composition in castor bean cotyledons and endosperm during early seedling growth.

JEFF LYNCH and ALICJA ZOBEL. Department of Chemistry, Trent University, Peterborough. Production of anthocyanins and other flavenoids in Acer saccharum and A. rubrum in response to stress.

FRIKILE NXUMALO and ALICJA ZOBEL. Department of Chemistry, Trent University, Peterborough. Influence of xanthotoxin, bergapten and imperatorin on oxygen consumption by plant and animal tissue.

MISBAH QURESHI, University of Toronto, Scarborough Campus. An immunocytochemical investigation to understand the biological function of meiotin-1 in Lilium longiflorum.

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S.E. RIVERS, P.S. SUMMERS and E.A. WERETILNYK, Department of Biology, McMaster University, Hamilton. Serine decarboxylase from spinach.

CHRIS SILVA and ALICJA ZOBEL. Department of Chemistry, Trent University, Peterborough. Ultraviolet radiation influences extrusion of phenolic compounds from alfalfa leaves.

D.D. SMITH, P.S. SUMMERS and E.A. WERETILNYK, Department of Biology, McMaster University, Hamilton. Light and salt regulation of a choline biosynthetic enzyme in spinach: s-adenosyl-l-methionine: phosphoethanolamine n-methyltransferase.

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## ABSTRACTS FOR TALKS

9:30

EVANS, RODGER C.\* and TIMOTHY A. DICKINSON. Department of Botany, University of Toronto, 25 Willcocks Street, Toronto CANADA M5S 3B2 and Department of Botany, Royal Ontario Museum, 100 Queen's Park, Toronto ON M5S 2C6 – Floral development of meristic variants in *Crataegus* sect. *Douglasii* Lindley (Rosaceae: Maloideae).

Meristic variation has been ascribed to differences in floral apex size. We have investigated this relationship in 20-stamen *Crataegus douglasii* var. *suksdorfii* and 10-stamen vars. *douglasii* and *rivularis*, all of which have similar-sized flowers at maturity. Developmental morphology was examined using scanning electron micrographs (SEM) of overwintering buds, morphometric analysis, and paraffin sections. The first floral organs to be initiated are five sepal primordia. Alternating with these, five additional primordia are initiated that appear to give rise to the petals, as well as to the members of five pairs of antesealous stamens. In var. *suksdorfii* further androecial development comprises the initiation of a second whorl of five antepetalous stamens, followed by the insertion of a stamen between the members of each pair in the first whorl, resulting in a total of 20 stamens. Floral development of the 10-stamen varieties differs from var. *suksdorfii* in that stamen initiation is completed with the first whorl. The relationship between primordium number and apex (area)\* (determined from SEM negatives) did not differ between 10- and 20-stamen varieties. Thus, in *Crataegus* sect. *Douglasii* meristic variation appears to be independent of floral apex size.

9:45

CHRISTINE M. KAMPNY, Department of Botany, University of Toronto, and Royal Ontario Museum, Toronto, Ontario. Variation in corolla development of Veroniceae (Scrophulariaceae).

Adult corollas of Veroniceae exhibit a diversity of shapes, largely due to differing proportions of tube and lobe lengths. Since the nearest relatives of the tribe have long corolla tubes, it is conceivable that all long-tubed corollas occurring within Veroniceae are ancestral. The homology of those tubes has been tested by measuring corolla tube and lobe growth throughout development, in one species each of 8 genera of Veroniceae, and one species of Digitaleae as outgroup. LOWESS plots of tube and lobe lengths relative to gynoecium length indicate that five developmental types of long corolla tube occur in the tribe, only one of which seems directly related to the ancestral type. Each tube type shows a differing pattern of heterochronic shifts during growth. Heterochronic shifts can help resolve the systematic placement of genera that are problematic because of convergent adult morphologies.

10:00

KITAGAWA, JUNKO\* and USHER POSLUSZNY. Department of Botany, University of Guelph, Guelph, Ontario, N1G 2W1 Canada.

- Preliminary study of inflorescence variations of *Lilaea scilloides*.

Small simple flowers like those formed in *Lilaea* may be primitive prefloral phases that came together to form common monocotyledonous flowers (Burger, 1977), or may be the result of secondary reduction and specialization. The arrangement and the number of flowers in inflorescences of *Lilaea scilloides* were examined to understand the evolution of the flowers of monocotyledons. Five kinds of flowers were observed. No variation of floral organ number in each flower type was seen. The flower number per inflorescences varied. Basal flower number was always 2 or 3 and seemed to influence the flower number in inflorescences. Both the occurrence and the position of bisexual flowers appeared to be at random. Certain numbers of flower per inflorescence seem to occur more frequently and the most common interval of increase of total flower number per inflorescence was 3. It's possible that the pattern of increase of flower number might tend to support Burger's hypothesis of monocotyledonous flower evolution.

10:15

Sage, Tammy L. Department of Botany, University of Toronto, 25 Willcocks Street, Toronto, Ontario M5S 3B2 -  
Ovarian Self-Incompatibility in *Asclepias exaltata*.  
Temporal and spatial aspects of self-incompatibility (SI) in *Asclepias exaltata* were characterized. No differences were noted between self-pollen and cross-pollen tube growth from the time of pollen tube germination to release of male gametes into the female gametophyte. Anatomical data indicates that the fusion of one male gamete with polar nuclei and fusion of a second male gamete to the egg nucleus occurs in both self- and cross-pollinations. Morphometric measurements of seed size indicate integument growth ceases 6 days following self-pollination but not cross-pollination. Analysis of endosperm and embryo development using sectioned material and ovule clearings indicate that endosperm and embryo growth is terminated by 6 days following self-pollination. Results indicate that *Asclepias* exhibits ovarian SI. Future studies on ovarian SI and the significance of ovarian SI in the evolution of angiosperms will be discussed.



10:30

CHROMOSOMAL DISTRIBUTION OF MEIOTIC MOLECULES

HASENKAMPE, C., JAYAWARDENE, N., QURESHI, M., AND DOOKHERAN, M.  
UNIVERSITY OF TORONTO, SCARBOROUGH CAMPUS

Homologous chromosomes come together during meiosis (synapse) and prepare for reciprocal genetic exchange. Three major steps of synapsis are homolog alignment, a check-for-homology, and synaptonemal complex (SC) formation between aligned homologs. Our research focuses on two distinct aspects of meiosis. The first area is elucidating the function of a class of DNA (zygDNA) with a possible role in the check-for-homology; the second area is the further characterization of a protein (meiotin-1) that appears to be important in determining meiotic chromatin structure. ZygDNA, is not replicated until after meiosis begins, and inhibition of this replication inhibits SC formation. Three putative zygDNA clones have been analyzed; all are members of different repetitive DNA families. Not all members of each family replicate during zygotene but there is evidence that a fraction of the members of two of the families may. We are currently investigating the chromosomal distribution of these sequences using *in situ* hybridization. --Meiotin-1, is a conserved, meiotic protein similar to histone H1, but is present, on average, only once every 5-13 nucleosomes. Its temporal and spatial distribution are consistent with its having a role in limiting chromosome condensation during prophase I. Using immunocytochemistry we are comparing the chromosomal distribution of meiotin-1 and histone H1, with respect to their location relative to the SC.

11:00

Gerrath, Jean M.<sup>1</sup> and Nancy G. Dengler<sup>2</sup> -- Primary vascular patterns in the Vitaceae. -- <sup>1</sup>Department of Horticultural Science, University of Guelph, Guelph, ON N1G 2W1 and <sup>2</sup>Department of Botany, University of Toronto, 25 Willcocks St., Toronto, ON M5S 3B2.

One of the oldest morphological questions is how to interpret the leaf-opposed tendril/inflorescence of the Vitaceae (grape family). To help answer this question, an anatomical analysis of primary vascular patterns in Cissus rhombifolia, Vitis riparia, and Leea guineensis, three taxa that represent the main architectural patterns found in the family, was carried out. Serial resin sections were reconstructed using computer, video, and camera lucida methods. The third method proved the most efficacious. The vascular pattern and connections of the tendril/inflorescence resemble those of the axillary bud most closely. However, we found no evidence to support the suggestion that the tendril/inflorescence represents a displaced axillary bud. Likewise, since the leaf and tendril/inflorescence vascular patterns differ, the evidence does not corroborate the hypothesis that the tendril represents the hypopodium of a laterally displaced leaf. Our results also failed to support a sympodial interpretation of the shoot, since tendril vascular development was delayed. Thus, based on previous developmental evidence and this study, we conclude that the present day vitaceous shoot is monopodial, but unique in its construction.

11:15

YOUNG, JANE P. Department of Botany, University of Toronto, Toronto, Ontario, M5S 3B2 Canada.

The developmental basis of heterophylly in *Ranunculus flabellaris*.

Variation in leaf form in response to changing environmental conditions (heterophylly) has been important in understanding the developmental basis of leaf shape modification. *Ranunculus flabellaris* exhibits drastic heterophylly upon changing water levels: terrestrial leaves have broad lobes, whereas underwater leaves have narrow elongate lobes. Plants submerged in a 25  $\mu$ M solution of ABA produce terrestrial-like leaves instead of submerged leaves. MorphoSys and multigroup principal component analysis were used to quantify leaf shape change emphasizing the stages of developmental divergence. Leaves developing under water are distinguished by both size, and to a lesser degree, shape, from terrestrial and ABA leaves. The smaller size of ABA leaves is expressed after Day 10, while terrestrial and underwater leaf differences do not occur until after Day 18. It is concluded that leaf determination in this species is a gradual process and can be influenced relatively late in development.

11:30

Daryl E. Enstone and Carol A. Peterson

Department of Biology, University of Waterloo, Waterloo, ON, N2L 3G1, Canada  
BAND PLASMOLYSIS AND THE DEVELOPMENT OF SUBERIN LAMELLAE  
IN THE EXODERMIS OF ZEA MAYS L. ROOTS

Corn is an example of a species with a uniform exodermis in which all cells comprising the layer are elongate and will develop both Casparian bands and suberin lamellae. Band plasmolysis, in which the plasmolyzed protoplast remains attached to walls with Casparian bands, is well documented in the endodermis. We find numerous examples of this phenomenon in maturing exodermal cells which have no suberin lamellae or are in the early stages of producing this layer. Thus, the Casparian band develops before the lamella and is tightly attached to the plasmalemma, as in the endodermis. These results are consistent with the function of the exodermis as a barrier to apoplastic ion movement. We have confirmed earlier reports that the exodermal development is patchy and normally occurs as a considerable distance from the root tip. Preliminary evidence indicates that exposure of part of a root to air induces the development of suberin lamellae in the majority of its exodermal cells. A root zone with a dead epidermis and almost complete exodermis should resist water loss to the environment but have a severely diminished capacity for ion uptake.



11:45

**Root growth under water stress.**

A. Darlington and J. Dat.

Faculty of Forestry, University of Toronto, 33 Willcocks St., Toronto. M5S 3B3

Root exploration and exploitation of the soil is a key factor in the plant's ability to survive drought. However the literature on conifer roots under drought is unclear with growth being either accelerated or reduced depending on the study. Humidity is presented as an alternative method of creating water stress within the plant. Dry atmospheric conditions lead to a significant reduction in the partitioning of biomass into the roots. More precisely, *in situ* monitoring of root growth of jack pine seedlings indicated the actual rate of root tip elongation was not reduced, instead there was a significant reduction in the initiation of new lateral roots under atmospheric drought. *In situ* monitoring of water potential indicated a large water potential gradient between the shoot and the roots which isolated the atmospheric drought to the shoot component. But, the gradient may also have reduced phloem translocation of assimilates out of the leaf, leading to the observed reduced root growth. The creation of the gradient may be largely dependent on the experimental droughting method, which may explain the range of root responses reported.

12:00

**POSSIBLE USE OF SCOPOLETIN AND IMPERATORIN AS ANTICANCER AGENTS**

Cheryl Ashby and Alicja Zobel, Dept. of Chemistry, Trent University, Peterborough, ON K9J 7B8

Coumarins and furanocoumarins are naturally occurring phenolic compounds found on the surfaces of many plants which are known to be protective agents for plant tissue. Mitotic indexes were evaluated after phases of mitosis had been counted under a light microscope using *Allium* root tips treated with scopoletin or imperatorin. One-hour incubations with 10 ppm scopoletin and 100 ppm imperatorin retarded mitosis, but after 24 hours imperatorin was not inhibitory. In the course of this study chromosomal aberrations were also counted. This study was able to demonstrate the possible use of scopoletin as an antimitotic agent, but imperatorin has not been found to be antimitotic.

2:15

UTILITIES OF A POLYCLONAL ANTISERUM DIRECTED AGAINST DNA  
BINDING PROTEINS FROM MEIOTIC CELLS

C. Daniel Riggs and Andrea Horsch. Department of Botany and Centre for Plant  
Biotechnology, University of Toronto, Scarborough College, 1265 Military Trail,  
Scarborough, Ontario M1C 1A4.

Our primary research interests center on the identification of proteins and genes specific to the process of meiosis. To design a molecular tool to dissect developmentally regulated gene expression, we have generated a polyclonal antiserum against proteins which bind to DNA cellulose. Proteins were extracted from meiotic prophase nuclei by sonication in a low salt buffer and were then passed over a lily DNA-cellulose column. The bound proteins were eluted with 0.5M NaCl and were injected into a rabbit to produce the antiserum. Immunoblotting experiments showed that the antiserum recognizes many polypeptides. Immunoblots of total protein extracts prepared from vegetative cells and cells at various stages of meiosis revealed a number of distinct differences. Several proteins appear transiently, and the patterns obtained indicate that the antiserum recognizes several putative meiosis-specific or microsporogenesis-specific proteins. Thus the antiserum is a useful tool to identify stage-specific or tissue-specific immunoreactive polypeptides. It also provides a tool to screen for the presence of an immunoreactive protein in protein purification schemes. In this presentation, the partial purification of a 75kD protein by this technique will be presented. The protein first appears during early pachytene and continues to increase in concentration until tetrads are formed. It is not found in extracts prepared from the other meiotic and vegetative tissues, and it is a low abundance protein in that it is not readily distinguishable on stained gels. The protein likely serves a role in microsporocyte development.

2:30

Kallie Keith\* and Peter McCourt  
Department of Botany, University of Toronto  
25 Willcocks St., Toronto, ON M5S 3B2

Molecular studies of late embryogenesis and seed development have emphasized differential gene expression as a means of identifying discrete stages of embryogenesis. Little has been done to identify factors that regulate the length of a given developmental stage or the degree of overlap between adjacent developmental programs. We designed a genetic screen to identify mutations that disrupt late embryo development without loss of hormonal responses. One such mutation, *fusca3* (*fus3*), alters late embryo functions, such as the establishment of dormancy and desiccation tolerance, and reduces storage protein levels. *fus3* cotyledons bear trichomes, and their ultrastructure is similar to that of leaf primordia. Immature *fus3* embryos enter germinative development, and the shoot apical meristems develop leaf primordia before seed desiccation begins. The cotyledons resemble leaf primordia, yet retain some cotyledon characteristics; thus cotyledon and leaf specific functions are expressed simultaneously. Taken together, these observations are consistent with a heterochronic interpretation of the *fus3* mutation.

2:45

**Isolation of ABA supersensitive mutants in *Arabidopsis thaliana*.**

Sean Cutler and Peter McCourt

Department of Botany, University of Toronto.

Abscissic Acid (ABA) has been implicated in several aspects of plant growth and development. Stomatal closure, seed maturation, seed dormancy and drought tolerance are among the many processes it is believed to control or mediate. To dissect the roles and pathways of hormone action *in vivo*, plant geneticists often screen for mutations which fail to respond to exogenously applied hormone. An approach complementary to this is to screen for mutants which "over-respond" to the hormone. Using this approach we have isolated several mutations capable of conferring ABA supersensitivity to *Arabidopsis* seeds. One mutant isolated, T12W, is recessive and appears to have greater seed dormancy than wildtype. This phenotype is consistent with ABA's role in dormancy induction. This mutant appears to be caused by a T-DNA insertion which will facilitate future cloning of the gene.

*Alcohol Dehydrogenase 1* Encodes the Anther-Predominant ADH Polypeptide from *Petunia* "Mitchell". E. Foster Atkinson and J.N. Strommer. Department of Molecular Biology and Genetics. University of Guelph, Guelph, ON, N1G 2W1.

*Petunia hybrida*, the common ornamental petunia, produces two alcohol dehydrogenase (ADH) polypeptides from two *Adh* genes. Both ADHs are induced by hypoxia in seedling tissues and expressed during development in anthers; ADH1 is predominant in anthers due to its abundance in pollen, while ADH2 is the major polypeptide induced by hypoxia. In contrast, the anther-predominant polypeptide from *Petunia* "Mitchell", a diploid plant arising from anther culture of a hybrid between *P. hybrida* and one of its progenitors, *P. axillaris*, is not induced by hypoxia. We have identified the gene that encodes the anther-predominant ADH polypeptide from *P. "Mitchell"* in three ways: 1) the anther-predominant polypeptide from *P. "Mitchell"* has the same relative mobility, on native polyacrylamide gels stained for ADH activity, as ADH1, the anther-predominant polypeptide from *P. hybrida*; 2) genomic DNA isolated from *P. "Mitchell"* yields *Adh1*-hybridizing RFLPs which correspond to those from *P. hybrida*; 3) RNA isolated from anthers of *P. "Mitchell"* hybridizes with *Adh1*, but not *Adh2*, sequences from *P. hybrida*. Using the above methods, we have identified the anther-predominant ADH polypeptide from *P. "Mitchell"* as the product of *Adh1*. We plan to study why *Adh1* from *P. "Mitchell"* is not induced by hypoxia; it may be due to the absence of a functional trans-activator, presence of a trans-acting inhibitor or mutation of the *Adh1* promoter.

D. BARABÉ Institut de Recherche en Biologie Végétale Montréal H1X 2B2  
The concepts of open system and closed system in phyllotaxis.

The great majority of mathematical models of phyllotaxis have dealt with mature inflorescences. But, to better understand phyllotaxis, it is necessary to analyse also the development of composing elements during organogenesis. The young inflorescence of *Symplocarpus* has a globular form. Floral primordia appear in successive rows, in alternate position. In the early stages, floral primordia have a spherical form. Subsequently, when their volume increases, the flowers take on a irregular tetragonal form. The floral parts appear at this stage. It is not possible to determine the exact sequence of formation of floral primordia. The pentagonal or hexagonal form of the mature flowers can be explained by the development of floral parts and the constraint caused by the contact between primordia. There are two periods in the development of the inflorescence: (a) before and (b) after the initiation of floral parts. Each of these periods corresponds to different morphogenetic processes. To determine the phyllotactic pattern, we must analyse the first period, because it can be obscured by the growth of flowers. The proportion between length and width is not the same before and after the appearance of floral parts. This analysis permits us to develop the concepts of open and closed system in phyllotaxis. In a closed system (e.g. spadix) differentiation of the floral primordia occurs on a closed surface whose shape and area can change, but that can not be renewed. In an open system (e.g. shoot apex) the lateral appendages are produced in cyclical fashion. At every cycle, one sees the restructuring of the apex.

BEECROFT, PENNY E. and JOHN N.A. LOTT. Department of Biology, McMaster University, Hamilton, Ontario, L8S 4K1 Canada. - Leakage of mineral nutrients from imbibing pollen grains.

Neutron activation analysis (NAA) was used to quantify the amounts of K, Na, Mg, Cl, Ca and Mn leaked from imbibing pollen grains. Pollen from four angiosperm and three gymnosperm species, specifically birch (*Betula pendula*), corn (*Zea mays*), spinach (*Spinacia oleracea*), squash (*Cucurbita maxima*), spruce (*Picea abies*), Austrian pine (*Pinus nigra*), and Scots pine (*Pinus sylvestris*) was studied. Pollen of all species studied, except squash, leaked detectable levels of the six elements measured; squash pollen leaked detectable amounts of all elements except for Mg. In general, more K was leaked from pollen than any other element and Mn showed the lowest level of leakage. Comparisons of the amount of an element leaked to the quantity of that element present in dry pollen showed that leakage represents a substantial loss of mineral nutrient reserves. Elements present in dry pollen in higher concentrations were leaked in higher amounts but not necessarily in higher percentages of the amount originally present. Pollen of species with smaller diameter grains generally leaked more of a given element than did pollen of species with larger diameter grains.

#### **Humidity and the hydraulic conductance of *Phaseolus vulgaris*.**

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It is commonly believed that the water conducting system of plants grown under dry conditions will be very different from more succulent conditions. However, most of this reasoning is based upon a very small number studies. Inability to reliably and precisely drought stressing plants complicate the impact of long term stress on the water conducting system of plants. These complications can be reduced when water stress is generated through the use of humidity control.

*Phaseolus vulgaris* cv. *provider* plants were grown under either dry (1.5 to 2.0 kPa) or humid (0.3 to 0.8 kPa) diurnal humidity regimes for two weeks, in specially designed growth chambers. The hydraulic conductances of excised stem segments were then determined. Prior to this determination possible cavitations were removed from some stems by root pressure. The dry atmospheric conditions lead to the development of a water conducting system with a substantially lower conductance. Since these differences were present in the rehydrated stems, the changes in conductance likely reflect anatomical changes in the xylem and not simply reflecting the impact of cavitations.

## COMPETITION OF PINE SEEDLINGS AND GRASS UNDER WATER STRESS

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This study was developed to determine the morphological and physiological effects of grass on young jack pine seedlings in a controlled environment, in order to identify the features that indicate whether a seedling or group of seedlings can survive competition. Morphological measurements were made of root collar diameters and heights, and chlorophyll fluorescence, temperature was measured by infrared thermography, and water potential and glutathione levels were determined. Seedlings grown in grass appeared under more stress than those grown without, as seen in the chlorophyll fluorescence parameter P-T. This parameter is an indicator of the seedling's CO<sub>2</sub> fixation potential: the greater the difference between the fluorescence levels at P and T, the greater the fixation potential (Hawkins and Lister, 1985). Seedlings grown without competition had P-T values less than those grown with competition, indicating that the CO<sub>2</sub> fixation potential of the former is the greater.

## IN SITU HYBRIDIZATION WITH LILY MEIOTIC CHROMOSOMES

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A key event which occurs during meiosis is synapsis. This is a multistep process which involves (1) the alignment of homologous chromosomes, (2) check for homology, and (3) synaptonemal complex (SC) formation between bivalents. Synapsis is essential to reciprocal genetic exchange, and reciprocal genetic exchange is essential to proper segregation of homologous chromosomes at Metaphase I. For reciprocal genetic exchange to occur, there has to be a check for homology. It is believed that in lily, this checking is performed by a specific set of DNA molecules called zygoteneDNA (zygDNA). These DNA sequences constitute 0.2% of the lily genome and are distributed more or less uniformly along pachytene chromosomes in clusters of about 5Kb in length. ZygDNA is believed to have a role in synapsis based on two lines of indirect evidence: (1) its time of replication coincides with the time of synapsis, (2) zygDNA replication is inhibited using high levels of deoxyadenosine, synapsis is also inhibited. Our lab has received a putative set of zygDNA clones and these have been labeled with digoxigenin and used for *in situ* hybridization on lily chromosome spread preparations. Preliminary work at the light microscope level indicates that these clones hybridize to sequences located near to or associated with the SC. This is promising because we have predicted that sequences involved in the check for homology should be located where the chromatin loops attach to the SC since this structure is believed to stabilize the paired state and provide the structural framework for recombination. Future studies hope to take this work to the electron microscope level to closer examine the precise location of zygDNA relative to the SC.

## The Expression and movement of Wheat Streak Mosaic Virus Coat Protein in infected wheat.

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The movement of Wheat Streak Mosaic Virus (WSMV) from the first inoculated leaf and into the second leaf was monitored by external symptom development, *in vivo* labelling and immunocytochemistry. The 45kD coat protein and 66, 40 and 78kD virus-encoded proteins were *de novo* synthesized in 2 days post- inoculation (d.p.i.)- old 2<sup>nd</sup> leaves. Thus, virus infectious material has moved into the second leaf as early as 2 d.p.i. Immunolabelling studies showed that coat protein antigen localized in the nuclei and cell walls at 2 d.p.i. Virus particles and/or coat protein were localized specifically in the cell wall plasmodesmatal regions at 4 d.p.i. At 7 d.p.i., the antiserum reacted with the plasmodesmata, the virus-encoded cylindrical inclusions associated with them, and the virus particles in the cytoplasm. Based on these results, we suggest that WSMV coat protein is involved in WSMV short- distance transport. Currently, nuclei are being isolated to verify the presence of the coat protein in the nuclei of the 2 d.p.i. infected tissue.

Development of leaf tendrils in pea: a progress report. Gerrath, Jean M., Joan E. Krochko and Judy N. Strommer. Department of Horticultural Science, University of Guelph, Guelph, ON N1G 2W1.-- A number of morphological mutations have been characterized in pea (*Pisum sativum* L.) that alter the form of one or other of the leaf components (stipule, leaflet, or tendril); one of these, *afila*, causes a replacement of leaflet primordia by a tendril complex. This report compares the development of lateral primordia in leaves from *Af* and *af* in an isogenic line (NLEP) using paraffin-embedded and sectioned material. Sections of the *Af* leaves were examined for planes and positions of cell division of the first order lateral primordia. Leaflet and tendril primordia could not be distinguished until P<sub>4</sub> (four leaves below the shoot apex) when cell divisions in leaflet primordia of *Af* become localized as periclinal and oblique divisions in the areas that will form the blade margin and the midvein. In *af* such cell divisions do not become localized. The strategy for the isolation of the *Af* gene is based on the assumption that it specifies a rare transcript, and that there is little or no expression of its mRNA in the mutant (*af*) line. Messenger RNA from terminal vegetative apices (inclusive of P<sub>4</sub>/P<sub>5</sub>) of an *Af* and *af* isogenic line (NLEP stock) are being used to construct enriched cDNA libraries using a repetitive subtractive hybridization. The large number apices required for polyA<sup>+</sup> isolation are generated by means of serial decapitations which force the growth of axillary buds. Initial identification of the target cDNA (*Af*) amongst several candidates will be based on tissue-specific distribution of its corresponding mRNA and cosegregation of the genotype with RFLPs in the F<sub>2</sub> of an *Af* x *af* cross. Initial screening of two sets of isogenic lines (*Af* and *af*, NLEP and Alaska) using a lentil cDNA (C49) has confirmed the presence of persistent heterozygosity around this locus.

LOTT, JOHN N. A., M. MARCIA WEST, BEN CLARK and PENNY E. BEECROFT. Department of Biology, McMaster University, Hamilton, Ontario L8S 4K1 Canada. - Changes in globoid crystal composition in castor bean cotyledons and endosperm during early seedling growth.

The endosperm and cotyledon tissues of *Ricinus communis* seeds and young seedlings were examined for changes in the mineral nutrient composition of globoid crystals during early seedling growth. The effect of providing mineral nutrients to developing seedlings on globoid crystal composition was also investigated. Globoid crystals in endosperm and cotyledon tissues of castor bean seeds contained P, Mg and K, as well as, trace levels of Ca, Fe and Zn. Irrespective of the addition of mineral nutrients, K levels in globoid crystals of endosperm and cotyledon tissues declined significantly during initial seedling growth. If mineral nutrients were not supplied during seedling growth, P and Mg globoid crystal levels increased significantly. Therefore, seedlings grown without mineral nutrients contained higher levels of P and Mg in globoid crystals of endosperm and cotyledon tissues than both dry seeds and seedlings grown with mineral nutrients. During early seedling growth, levels of Fe, Zn and Ca increased in cotyledon globoid crystals. Ca levels in globoid crystals of endosperm tissues also increased. The changes in Fe, Zn and Ca globoid crystal levels were not influenced by providing mineral nutrients to growing castor bean seedlings.

PRODUCTION OF ANTHOCYANINS AND OTHER FLAVONOIDS IN ACER  
SACCHARUM AND A. RUBRUM IN RESPONSE TO STRESS

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Flavonoids have long been perceived as compounds contributing merely to plant pigmentation; however, the current interest in medical applications of flavonoids as antioxidants raises new questions about the role of these compounds in the plant. The UV absorption spectra of the surface and interior flavonoids of various autumn leaves after UV irradiation were compared. Qualitative and quantitative differences in flavonoids, both on the surface and in the interior, were dependent on stress conditions, suggesting their role in plant defense response. We postulate as well that they can react as antioxidants in both the vacuole and outside the cells.



INFLUENCE OF XANTHOTOXIN, BERGAPTEN AND IMPERATORIN ON  
OXYGEN CONSUMPTION BY PLANT AND ANIMAL TISSUE  
Fikile Nxumalo and Alicja Zobel, Department of Chemistry,  
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Measurements of oxygen consumption were performed on mitochondrial particles and meristematic cells of *Allium cepa* root tips. The inhibitory effects of furanocoumarins on catalase were investigated. Bergapten was the most inhibitory of oxygen consumption in both meristematic and parenchymal cells. All the coumarins interfered with catalase activity, and both antagonistic and synergistic effects were noted as a function of concentration. Spectrophotometric analysis confirmed that at low concentrations these compounds may stimulate respiration. Results suggest that these furanocoumarins can stimulate or inhibit components of oxidative phosphorylation and the electron transport chain.

**An Immunocytochemical Investigation to Understand the Biological Function of  
Meiotin-1 in *Lilium longiflorum***  
Misbah Qureshi, Scarborough Campus, University of Toronto

From previous studies, a meiosis-specific protein has been characterized as having a different nucleosomal distribution than that of histone H1. This protein, called meiotin-1 is first detected in premeiotic G1, it reaches its greatest concentration in the leptotene and pachytene interval and is present at reduced levels by metaphase I. The focus of this study is to further examine the distribution of meiotin-1 during pachytene in *Lilium longiflorum* anthers. A combination of whole mount spreading and immunocytochemical techniques have been used and observations have been made at the light microscope level. The results from this study have confirmed earlier findings that meiotin-1 is distributed along the entire length of the chromosomes and that this distribution of meiotin-1 is similar to that of histone H1. However, unlike histone H1, meiotin-1 is more concentrated in patches at five to seven distinct locations on well-spread chromosome preparations. On tight chromosome preparations, one or two closely spaced patches are observed. Similar to histone H1, meiotin-1 also has a uniform distribution along the width of the chromatin-synaptonemal complex ensemble and it is not preferentially located at the periphery of the chromatin or at the central region of the SC. These results along with earlier findings suggest that meiotin-1 may have a role in chromatin condensation during prophase I of meiosis. The so called patch-like appearance of meiotin-1 may indicate its involvement with the organization of the telomeres into the bouquet-like structure.

### **Molecular cloning of cDNAs encoding meiotin-1: a meiotic protein associated with strings of nucleosomes.**

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Meiotin-1 is a chromatin-associated protein which is found predominantly in meiotic prophase nuclei. It is distributed in an unusual fashion relative to other nucleosomal proteins. Chromatin fractionation experiments reveal that, on average, there is one molecule of meiotin-1 per 5 to 10 nucleosomes. This distribution may be important for meiotic chromatin structure and meiotin-1 may have a role in controlling chromatin condensation. Using an antiserum directed against the protein, a cDNA clone was selected and the cDNA was used to screen for additional clones. Of the two clones selected, one is truncated and one is apparently full length. Computer analysis shows that the meiotin-1 polypeptide begins with a region having homology to the central globular domain of histone H1. I propose that meiotin-1 arose from histone H1 during the course of evolution to serve a distinct role in meiotic chromosome organization. DNA gel blot analysis demonstrates that there are homologous sequences in a number of species including fungi (but not yeast), ferns, and both mono- and dicotyledonous plants. RNA gel blot analysis shows that expression is restricted to anthers in which meiosis is occurring, and there are no detectable transcripts in bulbs, petals, leaves, roots or stems. Interestingly, computer analysis indicates that the cDNA encodes a protein of about 35kD, yet the polypeptide found in lily meiotic cells migrates at 43kD. Since one of the clones is apparently full length, these data are consistent with the interpretation that meiotin-1 is post-translationally modified. To confirm the identity of the cDNA as encoding the meiotin-1 polypeptide, the cDNA was expressed in *E. coli*, and the protein so produced was used to generate an antiserum. The antiserum recognizes the 43kD protein in lily, gives a weak reaction at 35kD and a strong signal at 30/31kD in induced bacteria carrying the meiotin-1 cDNA. Thus, it appears that the cDNA does encode a protein component of meiotin-1. The difference between the mass of the cDNA encoded protein and the native lily protein is being investigated.

### **SERINE DECARBOXYLASE FROM SPINACH**

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In spinach, choline serves as a precursor for the compatible osmolyte glycine betaine (betaine). Under salinity or drought, enhanced betaine synthesis in spinach is accompanied by the increased activity of several critical enzymes in the pathway of choline synthesis from ethanolamine. In this study, the enzyme responsible for the synthesis of ethanolamine by the decarboxylation of serine has been examined in crude extracts of control and salinized spinach. Serine decarboxylase activity is detected in extracts of leaves but not roots. Enzyme reactions are carried out in a total assay volume of 150  $\mu$ l for 60 min at 30° C and  $^{14}$ CO<sub>2</sub> evolved is linear in assays with up to 400  $\mu$ g protein. The pH optimum for the reaction is between 7.0 - 8.0 and the highest rates were observed using Hepes buffer. Serine decarboxylase activity in dialysed extracts is not increased by incubation with the cofactor pyridoxal phosphate. This enzyme undergoes a modest, approximately 1.5 fold increase in activity in spinach salinized step-wise to 300 mM NaCl. Though modest, this increase is comparable to that observed for the enzymes involved in choline synthesis and choline oxidation to form betaine. Up-regulation of serine decarboxylase activity may be necessary to accomodate an increased requirement of ethanolamine for betaine synthesis under osmotic stress.

# ULTRAVIOLET RADIATION INFLUENCES EXTRUSION OF PHENOLIC COMPOUNDS FROM ALFALFA LEAVES

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The decreasing ozone layer and the increase in UV radiation reaching the earth's surface has raised questions as to the survivability of agriculturally important crops. We irradiated alfalfa leaves and field-grown plants with monochromatic UV of 254 or 266 nm. They survived under such stress for several days. We observed increased extrusion of fluorescent compounds to the surface, of which one was identified as scopoletin. Extracts of the surface of such plants had only a limited effect on human pathogenic bacteria, but displayed effective fungicidal properties.

## LIGHT AND SALT REGULATION OF A CHOLINE BIOSYNTHETIC ENZYME IN SPINACH: S-ADENOSYL-L-METHIONINE: PHOSPHOETHANOLAMINE N-METHYLTRANSFERASE

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Choline is required by all plants as a component of lipids and in many plants for the synthesis of glycine betaine or choline-O-sulphate. However, the pathway of choline biosynthesis appears to differ between plant species. In chenopods, phosphocholine (PCho) is synthesized by the sequential N-methylation of phosphoethanolamine (PEA) and then PCho is hydrolysed to choline. The first N-methylation is catalyzed by the enzyme S-adenosyl-L-methionine: PEA N-methyltransferase (PEAMeT). PEAMeT activity is readily detected in leaves of spinach grown under an 8 h day. When 4-6 week old plants are transferred to continuous darkness, PEAMeT activity gradually decreases to <10% of the maximal daily light level by 42 h. When plants exposed to 42 h dark are returned to light, PEAMeT activity increased 1.5 to 2-fold over 4 h with pre-dark levels of activity being restored by 24 h. However, if exposure to continuous light exceeds 24 h, PEAMeT activity eventually declines. PEAMeT activity has been shown to increase 1.5 to 2-fold in spinach grown under an 8 h light cycle and salinized stepwise or salt-shocked with 200 mM NaCl. If plants kept for 42 h in the dark are salt-shocked and then left in the dark or transferred to continuous light, PEAMeT activity only increases in those plants exposed to light. Thus light plays an essential role in the salt-induced increase in PEAMeT activity.

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## Development of asymmetry in the leaf of *Begoniaceae* seedlings.

The phenomenon of asymmetry in the *Begoniaceae* is still not fully understood. The present study is an attempt to describe the development in the first leaves of the plant. We followed the development of the first and second leaves throughout their growth using scanning electron microscopy. The data collected so far concern three species of *Begonia* that show different degrees of asymmetry and varied types of venation (palmate versus pinnate): *Begonia fagifolia*, *Begonia subvillosa* var. *leptotrichia* and *Begonia kellermanii*. These data show that the degree of asymmetry of the first leaves is not correlated with that of mature leaves. For instance, the first leaves of *B. fagifolia* when compared to that of *B. subvillosa* var. *leptotrichia*, is more visibly asymmetrical. This appears unexpected since adult leaves of *B. subvillosa* var. *leptotrichia* are more asymmetrical than those of *B. fagifolia*. We also have observed other differences between the three species, such as the shape of the initium at the beginning of its development (curved inward versus flattened) and the fact that, contrary to the adult leaves, primordia are not covered by the stipules in the first or the second leaves of either one of the species (except in the case of the second leaf of *Begonia kellermanii*). We are now investing the development of later leaves.



